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SEVENTEEN SPONTANEOUSLY OCCURING MUTANTS OF *DROSOPHILA ANANASSAE* FROM SRI LANKA

by

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Abstract : An attempt was made to isolate morphological mutants from wild populations of *Drosophila ananassae* from two locations close to Colombo in Sri Lanka to estimate the mutation load in these wild populations.

Seventeen mutants of spontaneous origin have been isolated. From eight recessive mutants, namely, marginal cell cross vein wing (*mcv*), marginal cell double cross vein wing (*mdcv*), demi wing (*dw*), reduced eye (*re*), brown eye(*bw*), marginal celless wing (*mcl*), semi wing (*sw*) and cut wing (*ct*) it has been calculated that the hetrozygous mutant load in *D. ananassae* in the wild populations in Sri Lanka is 0.29 per individual. The value is seen to be much lower than the value obtained for *D. ananassae* in a Pacific Island by Spencer (0.63 per individual). A dominant mutant, Curled wing (*Cu*), was also obtained from the wild. The mutants hooked leg (*hl*), net wing (*nt*), balooned semi wing (*bsw*) arose from the cultures of reduced eye (*re*), hooked leg (*hl*) and semi wing(*sw*) respectively. Two other mutants contracted wing (*cnt*), vesiculated wing (*vs*) arose from the culture of Curled wing (*Cu*) and notch wing(*n*) mutant arose from the culture of marginal cell less wing (*mcl*). These have not been fully tested. The mutants marginal cell cross vein wing (*mcv*), marginal cell double cross veins wing (*mdcv*), demi wing (*dw*), reduced eye(*re*), marginal cell less wing (*mcl*), semi wing (*sw*), balooned semi wing (*bsw*) and Curled wing (*Cu*) to our knowledge is being reported for the first time. Seven mutants are due to autosomal recessive genes, four are recessive sex linked mutants and only one is a dominant autosomal gene.

Introduction: *Drosophila ananassae* is the species of *Drosophila* commonly found in the South Asian and the South, East Asian regions of the world, and is the drosophild fly commonly found in Sri Lanka. Genetical studies of *D. ananassae* were started in earnest on a large scale in Japan in 1921. Previous to this Metz in 1916, started cytological studies of these species. These were classified as *D. carribbea*, Sturtevant, collected from Panama and Cuba. In

1921, genetical studies of *D. ananassae* were started with the first discovery of a mutant which was Curled wing, an autosomal recessive gene, as reported by Sturtevant. These early studies have been cited by Kikkawa and Moriwaki (1937, 1938). Recently Moriwaki and Tobari (1975) have described more comprehensively the mutants in *D. ananassae*.

2. **Materials and Methods :** Two stations in Sri Lanka namely, Gangodawila and Nawinna were selected for sampling *D. ananassae* once a fortnight for isolation of spontaneously occuring mutants. Ten wild males of *D. ananassae* were mated individually to virgin females of the laboratory bred wild stock in the ratio of one male to one female. The F_1 adults emerging from each vial were sexed and the males and females were kept separately. After three days F_1 males and females were allowed to intercross in separate vials, each containing one virgin female and two males. After emergence of the F_2 progeny from intercrossing of F_1 offspring, they were screened for morphological mutants. The F_2 adults were anaesthetized by using diethyl ether and observed under a stereo-microscope and carefully and systematically observed for morphological deviants. When such deviants were isolated they were tested genetically to study their mode of inheritance.

Once a suspected mutant type was observed attempts were made to obtain a pure breeding colony from it. From those pure colonies that were ultimately obtained, genetic crosses were made to study the mode of inheritance of the mutant gene.

3. **Results :** Seventeen spontaneously occuring mutants of *D. ananassae* have been isolated from the wild in this study. They can be described as marginal cell cross vein wing (*mcv*), marginal cell double cross vein wing (*mdcv*), demi wing (*dw*), reduced eye (*re*), hooked leg(*hl*), net wing (*nt*), brown eye (*bw*), marginal cellless wing (*mcl*), cut wing (*ct*), semi wing (*sw*), balooned semi wing (*bsw*), Curled wing (*Cu*), contracted wing (*cnt*), vesiculated wing (*vs*), notch wing(*n*), garnet eye(*g*), and dark antennae (*da*). Three double mutants brown eye and Curled wing (*bw Cu*), brown eye and contracted wing (*bw cnt*) and brown eye and marginal cell less wing (*bw mcl*) were also isolated in this survey.

3.1 **Recessive autosomal mutants :** Combined F_2 results of the parental cross P_1 and reciprocal parental cross P_2 of the seven recessive autosomal mutants namely, marginal cell cross vein wing (*mcv*), marginal cell double cross vein wing (*mdcv*), demi wing (*dw*), reduced eye (*re*), hooked leg(*hl*), net wing (*nt*), and brown eye (*bw*) are presented in Table I.

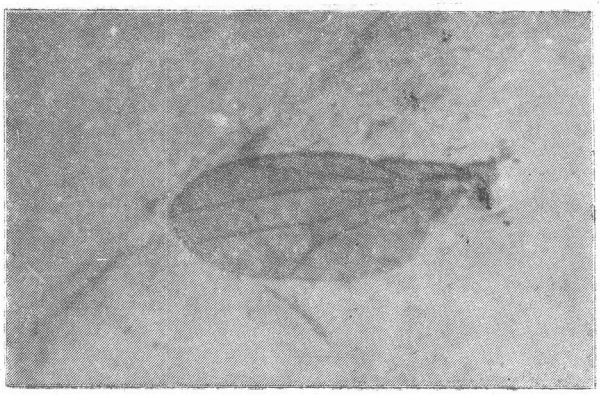

Plate I — *mcv* : marginal cell cross vein wing.
The extra cross vein is seen between the 1st and 2nd longitudinal veins.

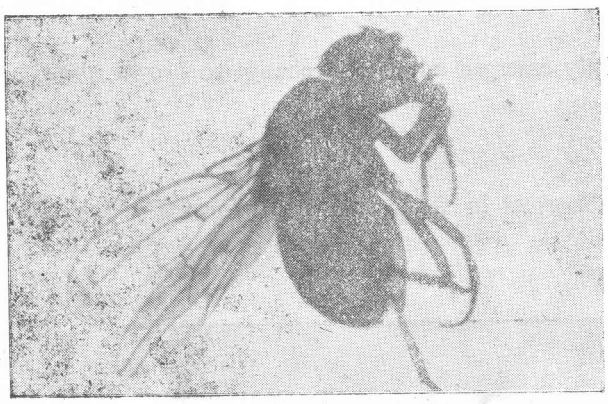

Plate II—*mdcv*: marginal cell double cross vein wing.

Except for the presence of an additional cross vein between L1 and L2 this mutant shows a similarlity to *mcv*.

Plate III — *dw* : demi wing.
The wing length is reduced to about half of the normal, while the breadth is about the same.

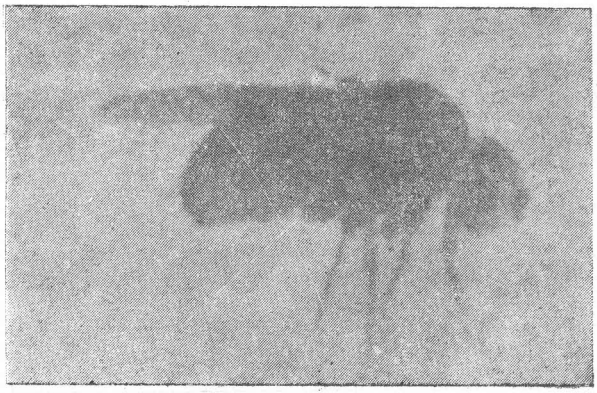

Plate IV — *re* : reduced eye.

Eye is smaller in size and is bar like than that of the wild type.

bw : brown eye.

The eyes are orange in newly emerged adults and change to brown about a day after emergence.

hl : hooked leg.

The tarsal segments are abnormal in shape and are deformed giving the appearence of a hook.

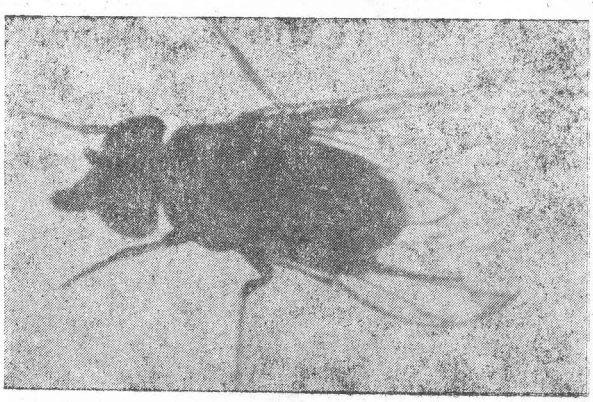

Plate V — *nt* : net wing.

The wings have a net work of extra veins at the tip.

3.2 **Recessive sex linked mutants:** The combined F_2 Results of the parental cross P_1 and the reciprocal parental cross P_2 of the four recessive sex linked mutants marginal cell less wing (*mcl*) semi wing (*sw*), balooned semi wing (*bsw*), and cut wing (*ct*) are presented in Table II.

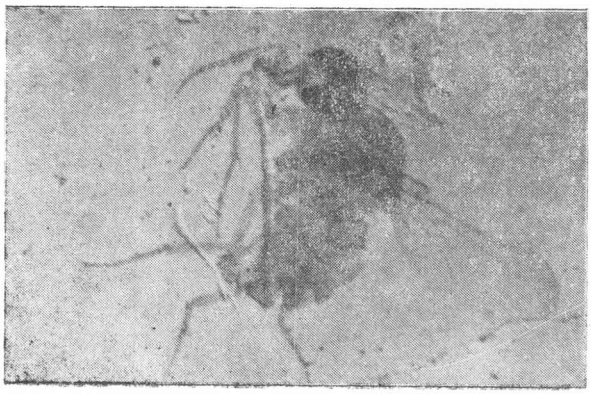

Plate VI — *mcl*: marginal cell less wing.

The marginal cell of the wings are completly missing

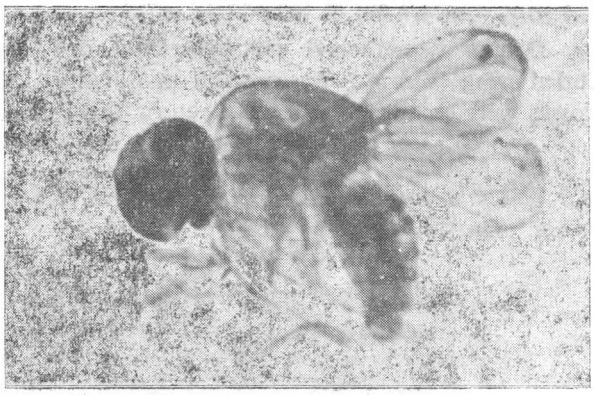

Plate VII — *sw* : semi wing.

Wing length varies from half to two thirds of the normal wing length.

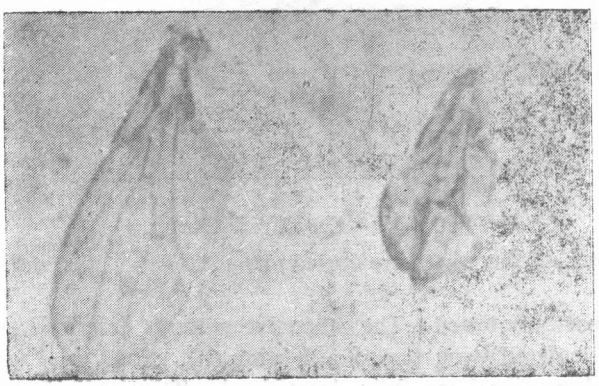

Plate VIII — *bsw* : balooned semi wing.

Except for the presence of a bubble in the middle of the wing this mutant is similar to the *sw* wing.

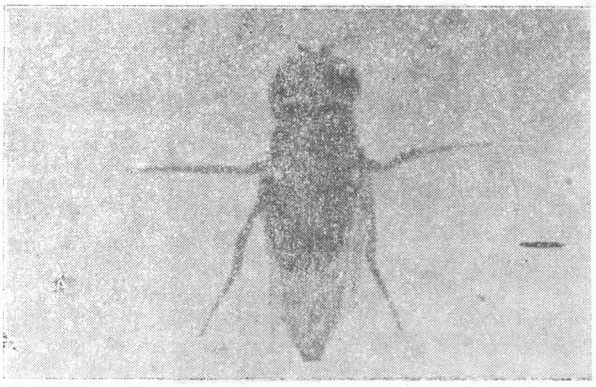

Plate IX — *ct* : cut wing.

The wing appears cut at the edges.

3.3 **Dominant autosomal mutants** : Only one dominant mutant namely, Curled wing (*Cu*) was observed in this study. The followiing experimental crosses were conducted and are given in Table III.

i. Cu/+ ♂♂ × +/+ ♀♀

ii. +/+ ♂♂ × Cu/+ ♀♀

iii. Cu/+ ♂♂ × Cu/+ ♀♀

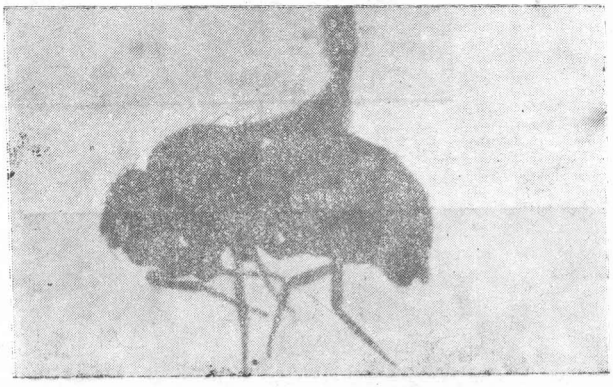

Plate X — *Cu* : Curled wing.

About two thirds of the wings are curled upward.

3.4 **Mutants not fully tested** : The other five mutants described as contracted wing (*cnt*), vesiculated wing (*vs*), notch wing (*n*), garnet eye (*g*), and dark antennae (*da*) have not been fully determined. They were lost before they could be tested.

cnt : contracted wing.

The wings are unexpanded and strongly turned upward.

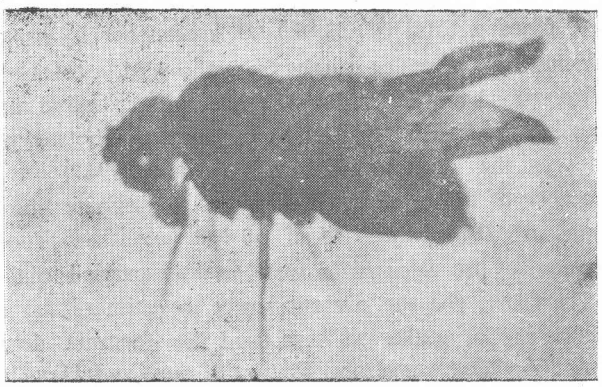

Plate XI — *vs* : vesiculated wing.

The wings are wrapped and wrinkled and they appear blistered and slightly turned upward.

These two may be phenotypes of the homozygous curled flies.

n : notch wing.

The terminal notches are present at the top of the wings.

4. **Discussion :** This is the first study of the mutants of *D. ananassae* from Sri Lanka. The recovery of naturally existing hetrozygous mutants from the wild populations of *D. ananassae* in Sri Lanka, have been used by us to also detect the mutation frequency in these populations.

Among these seventeen mutants the following nine mutants described as marginal cell cross vein wing (*mcv*), marginal cell double cross vein wing (*mdcv*), demi wing (*dw*), reduced eye (*re*), net wing (*nt*), marginal cell less wing (*mcl*), semi wing (*sw*), balooned semi wing (*bsw*) and Curled wing (*Cu*) are being reported for the first time for *D. ananassae*. It is seen that eye colour mutants and wing venation mutants seem to occur more frequently in Sri Lankan populations. The brown eye (*bw*), hooked leg (*hl*) and cut wing (*ct*) mutants have been reported previously in *D. ananassae* (Moriwaki and Tobari, 1975). As tests for allelism with these mutants have not been carried out with previously recorded mutants it is difficult to say whether the mutants isolated in this study are the same as that described by other workers; yet we have assigned the same symbols for them. The mode of inheritance of the remaining five mutants contracted wing (*cnt*), vesiculated wing (*vs*), notch wing (*n*), garnet eye (*g*) and dark antennae (*da*) could not be determined as they were lost before complete tests could be carried out on them.

The rate of mutation : In this survey, seventeen spontaneously occuring morphological mutants were isolated from progeny obtained from wild collected males of *D. ananassae*. Sixty two wild males from Nawinna produced 28,964 F_2 flies and fifty seven wild males from Gangodawila produced 24,855 F_2 flies.

Therefore, from a total of 53,819 *D. ananassae* F_2 progeny obtained from 119 pair matings, of males caught from the wild, a total of seventy three mutations were recorded. Sixty five of them were recessive mutants. These recorded recessive types can be used to estimate the hetrozygous frequency of these genes in the wild populations. Spencer (1947) has reviewed the work of a number of workers who have tried to estimate mutation loads in the populations of *Drosophila.* Most of the recent work on estimating mutation frequencies in *Drosophila* were mainly carried out for induced mutations. Therefore, for the present study to estimate the mutation load in *D. ananassae* in Sri Lanka we have used the formula devised by Craig and Hickey in 1966 which was a modification of the VandeHey (1964) equation. Our results indicate that the naturally occuring mutation frequency obtained for *D. ananassae* in Sri Lanka populations is 0.29 per individual. If the five lost abnormalities were also considered as true mutants the value would have been 0.32 per individual. The mutation frequency estimated for *D.ananassae* in Sri Lanka in the present study(0.29 per individual)seems to be much lower than that estimated for *D. ananassae* in the Pacific Islands by Spencer (0.63 per individual). The mutation frequency as calculated for *D. melanogaster* was 0.7 per individual which is also much higher than that obtained by us for *D. ananassae*. The estimated value for populations of *D. hydei* is 2.38 which is the highest value so far obtained for a mutation load for *drosophilids*.

The reason for the mutation frequency in Sri Lanka to be so low compared to Spencer's estimate for a Pacific Island may be due to either differences in screening methods or due to actual differences in the mutation load carried in these two populations created by biological factors.

The low frequency of mutations that we have obtained may have been also due to reasons adduced by VandeHey (1964). He has suggested that the chance of successful recovery of recessive mutants depends on the number of F_1 sibmatings which produced progeny for examination. In the present study only ten or less than ten F_1 matings were selected from each wild collected male. Therefore, some mutants may have been missed out or some additional mutations may have appeared in later generations. Also some pair matings produced very few offspring without any mutant flies.

Table 1—F_2 results of the parental crosses, P_1 and P_2 of seven recessive autosomal mutants of *D. ananassae* in Sri Lanka

Mutant and cross	*Wild type* ♀♀	♂♂	*Total no of Wild type*	*Mutant type* ♀♀	♂♂	*Total no of mutant type*	X^2 *Value*
(1) Marginal cell cross—vein wing (mcv)							
(*a*) mcv/mcv ♂♂ X +/+ ♀♀	303	271	574	123	173	296	37.78**
(*b*) mcv/mcv ♀♀ X +/+ ♂♂	145	157	302	33	77	110	0.64
(2) Marginal cell double cross vein wing (mdcv)							
(*a*) mdcv/mdcv ♂♂ X +/+ ♀♀	158	190	348	51	86	136	2.48
(*b*) mdcv/mdcv ♀♀ X +/+ ♂♂	192	189	381	81	57	138	0.70
(3) Demi wing (dw)							
(*a*) dw/dw ♂♂ X +/+ ♀♀	551	457	1008	146	134	280	7.30**
(*b*) dw/dw ♀♀ X +/+ ♂♂	136	132	268	22	28	50	14.60**
(4) Reduced eye (re)							
(*a*) re/re ♂♂ X +/+ ♀♀	466	346	812	134	107	241	2.51
(*b*) re/re ♀♀ X +/+ ♂♂	588	451	1129	196	178	374	0.02
(5) Brown eye (bw)							
(*a*) bw/bw ♂♂ X +/+ ♀♀	421	435	856	148	119	267	0.89
(*b*) bw/bw ♀♀ X +/+ ♂♂	508	494	1002	156	161	317	0.66
(6) Hooked leg (hl)							
(*a*) hl/hl ♂♂ X +/+ ♀♀	165	152	317	27	36	63	14.37**
(*b*) hl/hl ♀♀ X +/+ ♀♂	256	276	532	30	43	73	53.98**
(7) Net wing (nt)							
(*a*) nt/nt ♂♂ X + ♀♀	129	121	250	35	26	61	4.81
(*b*) nt/nt ♀♀ X + ♂♂	124	114	238	19	28	47	11.00**

**Highly significant

Table II—F_2 results of the parental crosses, P_1 and P_2, of four recessive sex- linked mutants of *D. ananassae* in Sri Lanka

Mutant and cross	*Wild type* ♀♀	♂♂	*Total no of wild types*	*Mutant type* ♀♀	♂♂	*Total no of mutant type*	X^2 *Value*
(1) Marginal cell less wing (*mcl*)							
(*a*) mcl/y ♂♂ X +/+ ♀♀	2369	1270	3039	—	1198	1158	5.96
(*b*) mcl/mcl ♀♀ X +/*y* ♂♂	235	239	474	200	227	427	4.10
(2) Semi wing (*sw*)							
(*a*) sw/y ♂♂ X +/+ ♀♀	419	181	600	—	126	126	25.61**
(*b*) sw/sw ♀♀ X +/*y* ♂♂	70	63	133	45	51	96	6.72
(3) Balooned semi wing (*bsw*)							
(*a*) bsw/y ♂♂ +/+ ♀♀	312	165	477	—	98	98	19.97**
(*b*) bsw/bsw ♀♀ X +/*y* ♂♂	68	51	119	34	40	74	13.86
(4) Cut wing (*ct*)							
(*a*) ct/y ♂♂ X +/+ ♀♀	564	238	802	—	264	264	4.87
(*b*) ct/ct ♀♀ X +/*y* ♂♂	280	292	572	190	246	436	24.86**

**Highly significant

Table III — F_1 Results of Curly wing, dominant autosomal mutant of *D. ananassae* in Sri Lanka

Mutant and cross	*Wild type* ♀♀	♂♂	*Total no of wild type*	*Mutant type* ♀♀	♂♂	*Total no of mutant type*	X^2 *Value*
(1) Cu/+ ♂♂ X +/+ ♀♀	321	257	578	290	246	536	1.58
(2) +/+ ♂♂ X Cu/+ ♀♀	253	241	494	217	174	391	11.99**
(3) Cu/+ ♂♂ Cu/+ ♀♀	204	219	423	487	43	926	2.03

** Highly significant

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